Computer Simulations of the Electric Interactions between the Phospholipid Head-Groups and Ionic Admixtures in the Membrane Surface

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Z. Naturforsch. **56 c,** 402–406 (2001); received March 3, 2000/January 24, 2001

Phospholipid Liposomes, Dipole Matrix, Binding Energy

Some phospholipids (e.g. lecithin) form a system of electric dipoles on the membrane surface layer. In the case of lecithin the positive dipole charge is located on the choline and the negative one on the phosphoric molecule group. These dipoles are arranged almost parallel to the membrane surface. Taking the dipole membrane structure as a base for further investigations, a computer model of the electrostatic interaction between the dipole system and the ionic admixture was investigated. The model presumes hexagonal centered or a rectangular flat geometry of the 121 dipoles distribution. The dipoles may rotate freely around round the motionless symmetry axis perpendicular to the system surface. The initial state is given by fixing the geometry of the dipole matrix and ionic admixture distribution. Subsequently this system underwent a computer simulation which consisted of a calculation of resultant force moments acting on each dipole caused by other dipoles and ions. These force moments lead the system to the equilibrium state (minimum of the binding energy). The minimum energy value of the dipoles system depends on concentration and charge of the admixtured ions. The results of repeated simulations indicate that the system achieve the least of all binding energy (the most stable equilibrium state) at 1.5% concentration of admixtured ions in case of ion charge equal to 1Q (where Q denotes arbitrary unit of ion charge) and at 2.5% concentration of admixtured ions in case of ion charge equal to 2Q. The calculated results are in a good agreement with the experimental.

Introduction

Physical and biological properties of the lipid bilayer, which constitutes the structural core of biological membranes, may be modified by introducing biologically active substances into their structure. For instance, studies conducted so far indicate that quaternary ammonium salts (AS) (those belonging to biologically active compounds) increase the fluidity of the liposome membranes (Podolak *et al.*, 1987, 1989; Podolak, 1991; Subczyński *et al.,* 1988) and the permeability of the membranes is increased by sulphate ions (Kuczera *et al.*, 1987, 1989). Moreover, it is known that presence of AS causes destruction of biological membranes (Kleszczyńska *et al.*, 1986; Witek and Grobelny, 1978; Cubkova *et al.*, 1981; Lindstedt *et al.*, 1990; Balgavy and Devinsky, 1994) as well as black lipid membranes (BLM) (Sarapuk *et al.*, 1986). These phenomena were observed at a relatively high concentration of the compounds studied. At a low concentration of both AS and other biologically active compounds, a bimodal effect of their action on biological membranes (Isomaa *et al.*, 1989) as well as on model membranes (Podolak *et al.*, 1996) was observed. This means that up to some critical compound concentration the membrane structure becomes more and more stable and rigid but after exceeding this value, the biological membranes undergo destruction (they increase the speed of the erythrocyte hemolysis) and the lipid membranes increase their fluidity. Podolak *et al.* (1996) reported a hypothesis that electrostatic forces occurring on the membrane surface layer are responsible for the AS – induced bimodal effect. Many of the membrane lipid molecules (like lecithin) contain electrically charged groups in their polar heads that give them an electric dipole feature. The active admixtures (e.g. AS) are often amphiphilic ions. At a low concentration these ions may arrange lipid polar heads around themselves, thus increasing the binding energy of the membrane surface layer. However, when the ion concentration exceeds a certain value, it may be assumed that the destabilizing factor of the membrane structure will predomi-

0939–5075/2001/0500–0402 $ 06.00

nate, which is caused by electrical repulsion forces between these ions.

The objective of this work was to compose a computer model that could simulate the electric interactions in the surface membrane layer. The layer in the model was represented by a dipole system constituting the added regular and flat matrix ionic admixtures. The presented computer model allowed to visualize the process how the equilibrium state of the dipole systems was achieved and to calculate the binding energy of these states.

Computer Model

Accepting the dipole structure of the membrane surface as a base, a computer model of the electrostatic interaction between polar head – groups (represented by the dipole system) and ionic admixture was elaborated. The model presumes the hexagonal centered or rectangular flat geometry of the dipole distribution. Dipoles may rotate freely round the motionless symmetry axis perpendicular to the system surface. The initial state is given by setting the geometry of the dipole matrix and ionic admixture distribution. The system defined in this way, is then influenced by a computer simulation, which gradually changes the orientation of the dipoles according to the resultant moment of the force acting on each of the dipoles but arising from other dipoles and ions of the system. The simulation is repeated until the equilibrium state (minimum of the binding energy) is obtained. Two electric dipole matrixes were used. The rectangular one consists of two-dimensional cells with the nodes composing a square (Fig. 1a) and the hexagonal centered one consists of cells with the nodes composing a centered regular hexagon – (Fig. 1b) (an elementary cell of the matrix is a centered regular hexagon). The hexagonal centered structure of the polar head groups of the lecithin surface membrane layer was introduced, among others, by Raudino and Mauzerall, (1986). In both cases the rotation axis of the dipole is fixed in the matrix nodes, and the distance between them is twice the dipole length (following Barrat and Laggner, 1974). The size of the matrix is determined by the number of nodes introduced into the computer at the beginning of the simulation process.

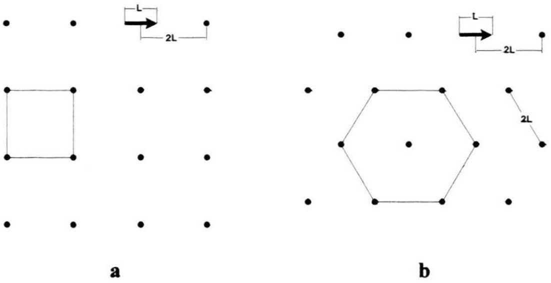

Fig. 1. Patterns of the dipole systems applied in the computer simulation. (a) – rectangular matrix with an elementary cell marked, (b) – centered hexagonal matrix with an elementary cell marked, L – dipole length, 2L – distance between matrix nodes.

At start of the simulation, the computer application used gives a choice of the following three types of dipole arrangement for the chosen matrix: parallel – all the dipoles are oriented the same direction, antiparallel – dipoles in the next row are oriented in opposite direction, random – dipole angles are at random (generated by the random number generator).

The application of an ionic admixture into the matrix is carried out by manual pointing (using a mouse) of the matrix node, where the charge is to be placed. The value of the admixture charge is typed with a keyboard. After all the parameters are set, the main simulation procedure starts.

The simulation algorithm is based on three continuously repeated steps.

In the first step, the resultant moments of the force, acting on each dipole and originating from the rest of the elements (dipoles and ions), are calculated. Because of the small distance between the dipoles in comparison with the dipole size, the concept of the dipole moment cannot be used in the calculations. For this reason the moments of force are calculated directly by determining the sums of the force acting on each charge of a dipole separately.

The Coulomb electrostatic interaction $F = k(q_1q_2)/r^2$ in this model is assumed, where q_1 is the dipole charge, and q_2 the ion admixture charge (or any other dipole charge) with distance r from charge q_1. In the second step of the simulation, dipoles rotate with an angle $\Delta\alpha$, according to the calculated moment of force M. The rotation angle (when the angular velocity equals zero), is directly proportional to the moment of force M acting on this dipole, $\Delta\alpha = WSP * M$. The proportionality factor WSP denotes $1/2 * dt^2/J$, where J is the moment of inertia of the dipole and dt is the simulation time interval. The WSP factor is chosen experimentally in such a way that for the maximum possible value of the force moment M, acting in the system, the rotation angle $\Delta\alpha$ is small enough to prevent the system from crossing over to the equilibrium state, on the one hand, and big enough to achieve the equilibrium state as soon as possible on the other hand. To avoid oscillation of the dipole around the equilibrium position, it is assumed that its environment causes very strong vibration damping (the overdamping motion). Thus, at the beginning of each simulation step the initial angular velocity of each dipole is zero. In the third step of the simulation, the electric potential energy of the system is calculated (the binding energy). The value obtained is then compared with the previous one: if there is a noticeable difference, the whole process starts from the first step again. The simulation reaches an end when the system achieves the equilibrium state (successive steps of the simulation do not change the binding energy of the system).

Results and Discussion

The final results of this investigation are binding energies (E) of the dipole system, containing ionic admixtures at different concentrations and different values of the charge, calculated in the process of the computer simulation of the electric interaction occurring in this system.

Figs 2 A, 2B and corresponding Tables IA, IB, respectively, present the values of relative binding energy (E/E_0), calculated in relation to the energy of the dipole system without ionic admixtures (E_0). Relative energy values are positive; nevertheless, energies E and E_0 are negative. Accordingly, the increase in relative energy (E/E_0) corresponds to a decrease in binding energy (E).

Fig. 2A presents the relative energy of the system of 121 dipoles, whose center of symmetry is allocated in the rectangular matrix, depending on the concentration of the ionic admixture with a charge equal to 1Q and 2Q (arbitrary unit), for antiparallel or random dipole settings at the beginning of the simulation process. The figure shows that the dipole system with an admixture of ions

Table I. Comparison of the relative values of the binding energy of system E/E_0 depending on the number and concentration of the admixture ions, for admixture charge equaling $2Q$ and $1Q$, respectively. At the start of the simulation, the dipole system setting was antiparallel and random (A) – rectangular matrix, $E_0 = -41.997$ in the arbitrary unit and (B) – centered hexagonal matrix, $E_0 = -44.053$ in the arbitrary unit.

	Number of Ions	Ion Concentration C[%]	Relative energy of the system E/E_0			
			Ionic charge 2Q		Ionic charge 1Q	
			ANTIPARALLEL	RANDOM	ANTIPARALLEL	RANDOM
	0	0.00	1.000	1.000	1.000	1.000
	1	0.83	1.138	1.136	1.017	1.012
A	2	1.68	1.182	1.204	*1.027*	*1.023*
	3	2.54	*1.199*	*1.233*	0.979	1.018
	4	3.42	1.138	1.174	0.933	0.991
	5	4.31	0.975	0.982	0.924	0.923
	6	5.22	0.676	0.677	0.793	0.842
	8	6.14	0.171	0.159	–	0.692
	9	8.04	–	–	0.337	0.337
	0	0.00	1.000	1.000	1.000	1.000
	1	0.83	1.133	1.135	1.012	1.010
B	2	1.68	1.161	1.203	*1.025*	*1.021*
	3	2.54	*1.163*	*1.231*	0.969	0.969
	4	3.42	1.123	1.172	0.934	0.950
	5	4.31	0.972	0.980	0.924	0.922
	6	5.22	0.676	0.676	0.790	0.841
	8	6.14	0.155	0.159	0.337	0.337

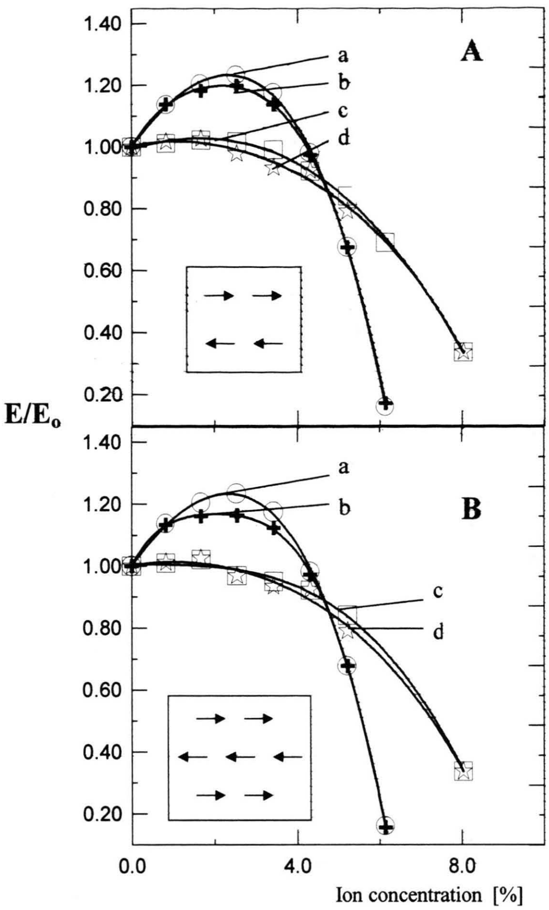

Fig. 2. Dependence of the relative value of the binding energy of the system equilibrium state E/Eo for rectangular (A) and hexagonal centered (B) geometry of the dipoles distribution on the ion admixture concentration. Symbol Eo denotes binding energy of the system without ions admixture (in the equilibrium state). (A) $E_o = -41.997$ and (B) $E_o = -44.053$ in the arbitrary unit.
a – Random angle distribution of the dipoles at the starting stage of the simulation process (charge of ion admixture is equal to 2Q – in the arbitrary unit);
b – antiparallel dipole distribution at the starting stage of the simulation process (charge of ion admixture – 2Q);
c – random angle distribution of the dipoles at the starting stage of the simulation process (charge of ion admixture – 1Q);
d – antiparallel dipole distribution at the starting stage of the simulation process (charge of ion admixture – 1Q).

of charge 1Q (curve c and d) reaches the extreme of relative energy E/Eo (equilibrium state) at about 1.5% admixture concentration. The value of the energy of the equilibrium state only insignificantly depends on the arrangement of dipoles at the beginning of the simulation. A slightly higher relative energy is achieved by a system with random dipole arrangement in comparison with the antiparallel one (curve c and δ in Fig. 2A). Fig. 2A also shows that the dipole system with an admixture of ions of charge 2Q (curve a and b) reaches the extreme of relative energy (*E/E*o) at about 2.5% admixture concentration, regardless of the initial arrangement of dipoles. Also in this case the system with a random dipole arrangement achieves a slightly higher relative binding energy than the antiparallel one. The difference in the energy extreme values, evoked by different settings of dipoles at the beginning of the simulation process, increased once a higher ion charge was introduced into the system (Fig. 2A).

Fig. 2B presents the dependence of the relative energy of the system of 121 dipoles, whose center of symmetry is allocated in the hexagonal matrix, on the concentration of the admixture of ions of charge 1Q and 2Q units for antiparallel or random dipole settings at the beginning of the simulation process. Similarly to the case of the rectangular matrix, the dipole system with an admixture of ions of charge 1Q reaches the extreme of relative binding energy *E/E*o at a concentration about 1.5% and 2.5% for ions of charge 2Q. This result does not depend on the arrangement of dipoles at the beginning of the simulation process (random or antiparallel). As in the case of rectangular matrix, at equilibrium state a slightly higher energy value, is obtained by a system starting with the random dipole arrangement in comparison with the antiparallel one. Data for Fig. 2B is shown in Table IB.

The findings obtained by computer simulations show that dipole systems of arranged in a flat, two-dimensional rectangular or centered hexagonal matrix with an admixture of ions, achieve the maximal (relative) binding energy at a specific (comparatively small) ionic admixture concentration. This concentration depends on the value of the ion charge introduced into the matrix. In case of the ion of charge 1Q, it is equal to about 1.5% and about 2.5% in the case of the ion of charge 2Q.

The results are qualitatively in agreement with the experimental investigations of interaction of organic ions with lecithin liposome membranes (Podolak *et al.*, 1996).

The appearance of the extreme in the binding energy of the dipole system at a specific ionic admixture concentration could be connected with aggregation of the dipoles around the ions. This process can strengthen and stiffen the system structure. At a higher ion concentration the process of dipole aggregation could be dominated by repulsion forces between the introduced ions. These forces may weaken the binding energy of the system when the ion concentration increases above the specific value. Both the dipole orientation at the beginning of the simulation process and the type of matrix do not affect substantially the binding energy obtained in the final equilibrium state. The research work carried out seems to confirm the hypothesis advanced by Podolak *et al.* (1996) about the dominant influence of the electrostatic interactions on the binding energy of the membrane components occurring on the surface layer of the liposome membranes. The fairly good agreement of the results obtained by entirely different initial dipole settings at the beginning of the simulation process gives the model applied in this study a more universal character and increases the credibility of the results.

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